

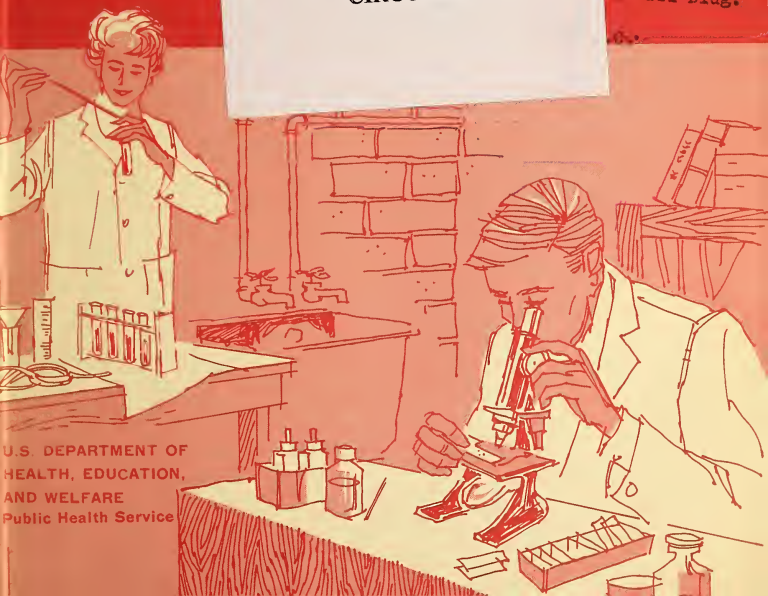
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*Health Mobilization Series*

F-4

# Laboratory Section of the Civil Defense Emergency Hospital

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U.S. DEPARTMENT OF  
HEALTH, EDUCATION,  
AND WELFARE  
Public Health Service

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# **Laboratory Section of the Civil Defense Emergency Hospital**

U.S. DEPARTMENT OF  
HEALTH, EDUCATION, AND WELFARE  
Public Health Service  
Division of Health Mobilization  
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# INTRODUCTION

In the event of a thermonuclear attack on the United States, a major proportion of existing hospital facilities would be destroyed. At the same time, there would be an unprecedented need for hospital beds and services. To help alleviate this problem, Civil Defense Emergency Hospitals (CDEH's) have been acquired by the Federal Government, loaned to States, and stored in communities throughout the country.

The CDEH is an austere, but functional, 200-bed general hospital designed to be set up in an existing structure, such as a school, armory, or other suitable building, as soon after attack as an area is safe from fallout. The unit can also be used to expand a permanent hospital. The CDEH contains supplies and equipment for the establishment of the following functional sections: Admitting and triage, operating rooms, wards, central supply, laboratory, pharmacy and X-ray. There are also provisions for auxiliary electric power and water supply.

Community health and civil defense leaders who are responsible for the storage of a CDEH unit are also responsible for preparing for its use. A staff should be selected and trained in the various aspects of setting up and operating the CDEH. By preattack planning and training, the community capability for coping with disaster can be increased.

The information contained in this booklet is intended to assist a community in preparing for the establishment of the laboratory section of the CDEH and in the use of the laboratory supplies and equipment provided with the unit for the performance of essential clinical laboratory procedures.

The first portion of this manual provides instructions for setting up the laboratory; assigning personnel; handling laboratory specimens, processing requests and reports; and general operational procedure.

The second portion provides detailed laboratory methods for performing selected tests in the categories of urinalysis, blood typing and grouping, hematology, blood chemistry, and bacteriology. The methods described are based on the complement of supplies and equipment contained in the Model 62 Civil Defense Emergency Hospital. A program is now under way to expand all older model CDEH's to this same capability by replacing the Laboratory Chest originally included in these models with a set of laboratory supplies and equipment to match those of the Model 62 CDEH.

The efficient operation of the CDEH laboratory unit requires pre-disaster assignment of individuals to staff the unit. Their responsibilities and functions must also be determined at that time. Participation by assignees in training and practice exercises is vital to the effective operation of the unit in a disaster situation. While the test methods included in this manual are intended to be those which are simplest to perform and most appropriate to disaster needs, it should be kept in mind that the proper interpretation of any laboratory finding is dependent upon the skill and experience of the laboratory worker performing the test. Accordingly, it is vital that bench personnel assigned to laboratory work be those with the maximum experience in performing clinical analyses or, at least, in performing related types of laboratory analyses.



# **ESTABLISHING THE LABORATORY SECTION**

## **A. SUPPLIES AND PROCEDURES**

The supplies and equipment of the laboratory section permit those laboratory procedures essential in diagnosis and treatment of the sick and injured in an emergency situation. A complete list of the individual laboratory items is included in Appendix A. Appendix B lists additional items needed in the laboratory which are stored in other sections of the hospital.

This manual describes only those tests essential to the provision of austere medical care in the early postattack period. Other tests, which may be judged desirable in view of particular operating conditions at an individual CDEH, can also be performed with the supplies and equipment provided. The scope of the laboratory can be further increased, particularly in the later postattack period, by the addition of equipment and supplies from other sources. At all times the policy pertaining to the scope of laboratory functions must be established by the chief of staff of the CDEH, with consideration given to the volume and type of workload, the number and skills of personnel available for laboratory work, and the availability of required supplies and equipment.

Staff members of those hospital sections requiring laboratory services should be familiar with the procedures for collecting and submitting laboratory specimens and for requesting and receiving laboratory reports. They should also be thoroughly aware of the limitations in laboratory capability and understand clearly that many routine laboratory services which are standard under normal conditions will not be available in a CDEH.

## **B. LOCATION AND FACILITIES**

The laboratory may be set up in any convenient location in the hospital, although it is advisable to place it as near as possible to those sections which will require its services (admitting and triage, wards, and operating rooms). Approximately 200 sq. ft. of floor space, preferably a separate room, should be assigned to the laboratory section. The area should be supplied with a laboratory-type workbench or an improvised substitute.

Also essential are a nearby sink with running water, a number of convenient electrical outlets and good lighting. A source of fuel for gas burners (gas line or bottled gas) is desirable but not absolutely essential to carry out the tests outlined in this manual.

Most of the supplies and equipment assigned to the laboratory are packed in wooden or heavy fiberboard boxes. After they are unpacked, these boxes can be used as improvised side tables or shelves. To complete the stock of laboratory essentials, as outlined in Appendix B, arrange to get the additional supplies from Central Supply or other appropriate sections of the hospital.

## C. PERSONNEL

The minimum personnel requirements for 24-hour laboratory operation (two shifts) are:

- two clinical laboratory technicians (or preferably one clinical laboratory technologist and one technician)

- two medical laboratory aides with laboratory experience (not necessarily clinical)

- two clerks

- two helpers

Predisaster, it is essential to recruit and train the best qualified volunteers available to staff the laboratory unit of the CDEH. Those technologists and technicians ordinarily employed in local hospitals, however, probably can not be counted on for staffing the CDEH as it is likely that in a disaster all fixed hospitals will be utilizing their entire complement of personnel for expanded operating capacity.

Predisaster training of the laboratory staff should include familiarization with the type of room in which the laboratory is likely to be set up and the arrangements necessary to convert it to a laboratory. Laboratory staff orientation should also include discussion of the limitations and difficulties which must be overcome in order to provide laboratory services.

Members of the laboratory staff also have the following specific duties:

### 1. Laboratory Technologists and Technicians

#### *Predisaster:*

#### a. Become familiar with:

- (1) kinds of laboratory services needed to support medical care

activities in a disaster.

(2) various items of equipment provided for the laboratory.

(3) operation of equipment for which they will be responsible. They should make arrangements for improvising or compensating for lack of items which ordinarily are available in fixed hospital laboratories.

b. Cooperate with the hospital leadership in determining laboratory staffing requirements.

c. Assist in the recruitment and training of laboratory personnel.

d. Participate in training programs and test exercises.

**Postdisaster:**

a. Report to chief of staff at operating site of the CDEH.

b. Supervise the setting up of the laboratory and laboratory operations. (One technician should be designated as chief of laboratory services if no technologist is available.)

c. Anticipate supply and personnel shortages and requisition additional supplies as needed.

d. Arrange a work schedule for the laboratory staff to permit 24-hour operation.

e. Perform or arrange for the performance of laboratory tests as requested by the medical staff.

f. Supervise the blood bank operation.

## **2. Clinical Laboratory Aides**

**Predisaster:**

a. Become familiar with:

(1) types of laboratory services likely to be required.

(2) equipment provided for the laboratory and the operation of that equipment for which they will be responsible.

b. Participate in training programs and test exercises.

**Postdisaster:**

a. Report to the laboratory chief at the operating site of the CDEH.

- b. Help move in and set up laboratory equipment and supplies as directed by the laboratory chief.
- c. Assist in the performance of laboratory tests under the supervision of a technologist or technician.
- d. Assist in other hospital medical care activities if the volume of laboratory test requirements decreases sufficiently.

### **3. Clerks**

#### ***Predisaster:***

- a. Learn laboratory terminology.
- b. Become familiar with the report and request forms to be used and the procedures for processing them.
- c. Participate in training programs and test exercises.

#### ***Postdisaster:***

- a. Report to laboratory chief at the operating site of the CDEH.
- b. Help move in and set up laboratory equipment and supplies as directed by the laboratory chief.
- c. Fill out report forms and supply request forms as directed by a technician.
- d. Perform simple laboratory procedures, if necessary, under close direction of a technician.
- e. Maintain laboratory record files.

### **4. Helpers**

#### ***Predisaster:***

- a. Become familiar with the equipment to be used in the laboratory.
- b. Participate in training programs and test exercises.

#### ***Postdisaster:***

- a. Help move in and set up laboratory equipment and supplies as directed by the laboratory chief.
- b. Wash and store laboratory equipment.
- c. Pick up and deliver supplies.
- d. Deliver laboratory reports and perform other errands as required.

D. FORMS

Three standard forms for requesting laboatory tests and for transmitting laboratory reports are packed with the Civil Defense Emergency Hospital:

- SF-514a (Urinalysis)
- SF-514b (Hematology)
- SF-514m (Miscellaneous Tests)

These standard forms (figs. 1, 2, and 3) are bound in sets of two, with carbon paper between. Both copies are forwarded to the laboratory by that hospital section requesting a test. The laboratory returns one completed copy of the form to the originating section upon completion of the test and files the other copy. A log of all laboratory tests requested should be maintained in the laboratory until the completed test reports are returned to the section which originated the request. The log should show the date of receipt, source and type of request, and a serial number.

In addition, an Emergency Hospital Supply Request Form (fig. 4) is recommended for requesting equipment and supplies from central supply or pharmacy. (It is not supplied with the CDEH.) This form is to be made out in triplicate; two copies are sent to the section from which supplies are requested and one is retained in the laboratory as a record of what was ordered. One copy is returned with the supplies ordered. If this form is not reproduced by the community for storage with the CDEH, a local hospital form or pads of unruled paper can be used as a substitute.

REGISTER OR UNIT NO.		WARD NO.	<input type="checkbox"/> REG. PATIENT
			<input type="checkbox"/> AMBULATORY
REQUESTED BY		DATE OF REQUEST	
DATE, TIME, AND METHOD OF COLLECTION			
PATIENT'S LAST NAME—FIRST NAME—MIDDLE NAME			
COLOR-APPEARANCE	MICROSCOPIC REMARKS		
REACTION			
SPECIFIC GRAVITY			
ALBUMIN			
SUGAR			
ACETONE			
BILE			
DATE OF REPORT	SIGNATURE (Specify Lab. if not part of requesting facility)		
NAME OF MEDICAL FACILITY			

Standard Form 514-A—Rev. June 1950.  
Bureau of the Budget Circular A-32

GPO: 1951-593079

URINALYSIS

Figure 1.—Urinalysis Form

PATIENT'S LAST NAME—FIRST NAME—MIDDLE NAME		REGISTER OR UNIT NO.		WARD NO.		<input type="checkbox"/> BED PATIENT <input type="checkbox"/> AMBULATORY	
		REQUESTED BY AND DATE					DATE COLLECTED
		CLINICAL DATA					
W.B.C.		R.B.C.					
DIFFERENTIAL COUNT		HEMATOCRIT					
NEUTROPHILES		HEMOGLOBIN					
BLASTS		BLEEDING TIME					
MYELOCYTES		COAGULATION TIME					
BANDS		BLOOD MORPHOLOGY; REMARKS					
LYMPHOCYTES							
MONOCYTES							
EOSINOPHILES							
BASOPHILES							
PLATELETS							
SEDIMENTATION RATE		DATE OF REPORT		SIGNATURE (Specify Lab. if not part of requesting facility)			
C.S.R.		NAME OF MEDICAL FACILITY					

Standard Form 314-B—Rev. June 1959,  
Bureau of the Budget Circular A-25

☆ GPO: 1961-860678

HEMATOLOGY

REGISTER OR UNIT NO.		WARD NO.	<input type="checkbox"/> BED PATIENT
			<input type="checkbox"/> AMBULATORY
REQUESTED BY AND DATE		DATE AND TIME COLLECTED	
CLINICAL DATA			
PATIENT'S LAST NAME-FIRST NAME-MIDDLE NAME			
SPECIMEN AND SOURCE		EXAMINATION REQUESTED	
RESULT			

DAYS OF REPORT	SIGNATURE (Specify Lab. if not part of requesting facility)
NAME OF MEDICAL FACILITY	

Standard Form 514-M-Ecc. June 1958. Bureau of the Budget Circular A-32	GPO c55-16-56808-6	MISCELLANEOUS
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Figures 2 and 3.—Hematology and Miscellaneous Text or Examination Report Forms

<h2 style="margin: 0;">EMERGENCY HOSPITAL SUPPLY REQUEST FORM</h2>		
FROM _____ SECTION _____	Date _____	
ROOM _____	Time _____	
<div style="display: flex; justify-content: space-between; align-items: center;"> <span>TO: (check one)</span> <div style="display: flex; gap: 20px;"> <span><input type="checkbox"/> STERILE SUPPLY</span> <span><input type="checkbox"/> PHARMACY</span> </div> </div> <div style="display: flex; justify-content: center; align-items: center; margin-top: 10px;"> <span><input type="checkbox"/> STORES</span> </div>		
QUANTITY	ITEM NEEDED	IDENTIFYING NO.
DISPOSITION OF REQUEST: <div style="display: flex; justify-content: space-around; margin-top: 5px;"> <span><input type="checkbox"/> ISSUED</span> <span><input type="checkbox"/> NOT AVAILABLE REORDER _____</span> <span><input type="checkbox"/> OUT OF STOCK</span> </div>		
INSTRUCTIONS: Prepare in triplicate. Send two copies to Sterile Supply, Pharmacy or Stores. Retain one copy.		

Figure 4.—Emergency Hospital Supply Request Form

# URINALYSIS

## A. COLLECTION AND PREPARATION OF SPECIMEN

Special wide-mouth bottles for the collection of urine samples are included in the laboratory supplies and equipment. These should be thoroughly cleaned and dried before delivery to the ward or other area where specimens are to be collected. Cardboard stoppers are also provided for distribution with the bottles.

A specimen is ordinarily collected directly into the bottle to avoid the possibility of contamination. The bottle is capped with the cardboard stopper on which has been written the name of the patient, the date and time in blanks provided. After completion of all required tests, the remaining portions of the specimen are discarded and the bottle rinsed, washed, and dried for reuse.

## B. TESTS

Tests should be performed on specimens as soon as possible after collection, as decomposition of urine occurs rapidly at room temperature. It should be remembered that urine may contain infectious organisms, such as those of tuberculosis, typhoid, or gonorrhea, and suitable precautions in handling and disposal of specimens and in cleaning up after laboratory work should be observed.

The reagents used in the tests described below should be handled carefully to avoid contamination and undue exposure to air, moisture, light, fumes, and heat. Bottles should be recapped tightly immediately after each use. Some of the reagents are especially subject to deterioration, and if the quality of any reagent is suspected, control tests on known samples should be performed to check its response and sensitiveness.

### 1. Acetone

#### a. Description

A color-comparison test for the presence of acetone, ketone bodies,

or acetoacetic acid in urine based upon the formation of a purple colored complex with sodium nitroprusside in the presence of aminoacetic acid.

**b. Equipment and supplies**

- (1) Acetone Test Tablets, with Color Chart.
- (2) Paper, Filter, Qualitative.
- (3) Pipette, Dropping (Medicine Dropper).

**c. Procedure**

- (1) Place one tablet on a small piece of filter paper, other white paper, or any clean surface.
- (2) Using a dropper, place one drop of the urine specimen on the tablet.
- (3) After 30 seconds compare the color of the upper surface of the tablet with the color chart.

**d. Evaluation**

*Negative:* No lavender or purple color appears.

*Positive:* A lavender or purple color appears, the depth of color depending upon the amount of acetone bodies present. Report the results as *trace*, *moderate*, or *strongly positive*, as determined by comparison with the color chart.

*Normal:* Negative.

*Sensitiveness:* A trace positive finding indicates approximately 10 mg. of acetone or acetoacetic acid per 100 ml. of urine.

**e. Additional information**

A definite yellow or orange discoloration of the reagent tablets or failure of the tablet to absorb the drop of urine sample within 30 seconds may be an indication of deterioration of the reagent tablets. The sensitiveness of any bottle of tablets showing such defects should be checked before reporting any results of findings with them.

**2. Albumin**

**a. Description**

A turbidity test for the presence of protein in urine based upon the precipitation of protein by sulfosalicylic acid.

**b. Equipment and supplies**

- (1) Albumin Test Tablets.



- (2) Bottle, Screw Cap, Prescription, 2 oz.
- (3) Cylinder, Graduated, Laboratory, 100 ml.
- (4) Funnel, Common, Laboratory, 65 mm.
- (5) Label, Gummed, Blank.
- (6) Paper, Filter, Qualitative.
- (7) Pipette, Serological, 5 ml.
- (8) Test Tube, 13 x 100 mm.
- (9) Water, Demineralized (or Distilled).

#### **c. Procedure**

- (1) Dissolved four reagent tablets in 30 ml. (1 fl. oz.) of demineralized water. This reagent solution is ready for use as soon as the tablets have dissolved and effervescence has subsided.
- (2) Put equal parts of reagent solution and the urine sample (2 ml. each) in a test tube and shake gently.
- (3) Immediately observe for any turbidity against a dark background.

#### **d. Evaluation**

*Negative:* No turbidity is produced.

*Positive:* A turbidity is produced, the degree of cloudiness depending upon the amount of protein present. Report the results as *trace*, *moderate*, or *strongly positive*, according to the degree of turbidity.

*Normal:* Negative.

*Sensitiveness:* A trace positive result indicates approximately 10 mg. of protein per 100 ml. of urine.

#### **e. Additional information**

- (1) If the original urine specimen is noticeably cloudy, it should be filtered or centrifuged before performing this test. If only a very slight turbidity is present in the sample, this operation may not be necessary as the untreated sample can be used as a control to determine if there is any increase in turbidity.
- (2) This test may falsely indicate positive results when the patient is receiving tolbutamide, certain X-ray media, or massive doses of penicillin.
- (3) The reagent solution prepared from the tablets can be kept

up to 30 days if it is stored in a tightly closed container in a dark place.

### 3. Bilirubin

#### a. Description

A color-comparison test for the presence of bilirubin in urine based upon the formation of a blue or purple colored compound with p-nitrobenzene diazonium p-toluene sulfonate.

#### b. Equipment and supplies

- (1) Bilirubin Test Kit (includes test tablets and fiber mats).
- (2) Pipette, Dropping (Medicine Dropper).

#### c. Procedure

- (1) Put five drops of the urine specimen on a square of the asbestos-cellulose mat supplied with the kit.
- (2) Place one reagent tablet on the center of the moistened area of the mat.
- (3) Flow two drops of water over the surface of the tablet, thus causing some of the tablet to dissolve and wash onto the mat.
- (4) Observe for 30 seconds for the formation of a blue or purple color on the mat around the tablet.

#### d. Evaluation

*Negative:* No blue or purple color appears on the mat within 30 seconds. (A slight pink or red coloration should be interpreted as negative.)

*Positive:* A blue or purple color appears on the mat around the tablet within 30 seconds. The amount of bilirubin present in the sample is roughly proportionate to the speed and intensity of the color reaction. Report a positive result as *trace*, *moderate* or *strongly positive*.

*Normal:* Negative.

*Sensitiveness:* A trace positive result indicates 0.05 to 0.1 mg of bilirubin per 100 ml. of urine.

#### e. Additional information

- (1) The tablets may darken from their original cream color on long storage, particularly at high temperatures, but this does not necessarily mean that they are unsuitable for use.
- (2) Atypical color reactions in this test may be observed when patient is receiving certain drugs, particularly urinary anti-infectives such as phenylazodiaminopyridine or ethoxazine.

## 4. Glucose

### a. Description

A color-comparison test for the presence of glucose (dextrose) in urine based on the oxidation of glucose by glucose oxidase to form gluconic acid and hydrogen peroxide. The latter then reacts with a catalyst-chromogen combination to produce a color change (yellow to green or blue).

### b. Equipment and supplies

Test Paper, Urine Sugar, with Color Chart.

### c. Procedure

- (1) Withdraw approximately  $1\frac{1}{2}$  inches of test paper from the dispenser and tear off against the cutting edge.
- (2) Dip one end of the paper strip into the urine sample, remove, and wait 1 minute.
- (3) Immediately compare the darkest area of the strip with the color chart on the dispenser. For values of 0.5% (+++) or more, allow 1 additional minute before making the final color comparison.

### d. Evaluation

*Negative:* The test strip remains yellow in color.

*Positive:* A green or blue color appears on the strip, depending upon the amount of glucose present. Report the semiquantitative result in terms of plus values and percent as given on the color chart.

*Normal:* Negative

*Sensitiveness:* Depending upon the exact conditions, 0.01% to 0.1% of glucose in urine will be required to produce definite positive results.

### e. Additional information

- (1) Misleading results may be obtained if patient is taking therapeutic doses of ascorbic acid.
- (2) Sugars other than glucose are not detected by this test.

## 5. pH (Acidity or Alkalinity)

### a. Description

A color-comparison test for the pH of urine based upon the color

of a mixed indicator in the presence of a specific hydrogen ion concentration.

**b. Equipment and supplies**

Test Paper, pH, with Color Chart.

**c. Procedure**

- (1) Withdraw approximately  $1\frac{1}{2}$  inches of test paper from the dispenser and tear off against the cutting edge.
- (2) Dip one end of the paper strip into the urine sample and remove.
- (3) After a few seconds compare the color of the wetted portion of the paper with the color chart on the dispenser.

**d. Evaluation**

*pH*: Report the pH value to the nearest whole number by determining the best color match with the chart.

*Normal*: 6 or 7.

*Sensitiveness*: The usable range of the test paper is from pH3 (red-orange) to pH9 (blue-green). The neutral color (pH7) is yellow-green. With care, pH value can be estimated to 0.5 unit if required.

**e. Additional information**

This test should be performed as soon as possible after collection of the urine specimen, because the sample will increase in alkalinity on standing.

**6. Specific Gravity**

**a. Description**

A determination of the specific gravity of urine compared with water by the use of a hydrometer (urinometer).

**b. Equipment and supplies**

- (1) Urinometer, Squibb, with Cylinder.
- (2) Paper, Filter, Qualitative.

**c. Procedure**

- (1) Transfer enough of the urine sample to the urinometer cylinder so that the float, when inserted, will float freely and not touch the bottom of the cylinder.

(2) Remove any foam or bubbles from the surface of the sample by touching with a strip of filter paper.

(3) Insert the float of the urinometer into the cylinder with a twist to prevent the adherence of air bubbles and to keep the float away from the sides of the cylinder.

(4) When the float is stationary and floating freely (not touching sides or bottom of cylinder), read the specific gravity from the scale on the stem of the float at the point indicated by the bottom of the meniscus of the liquid.

#### **d. Evaluation**

*Specific gravity:* Report the reading of the urinometer to 3 decimal places.

*Normal:* 1.005 to 1.030.

*Sensitiveness:* The specific gravity can be estimated to 0.001 or 0.002 unit with this instrument.

#### **e. Additional information**

For precise work the temperature of the specimen should be brought to the value for which the urinometer is calibrated (usually marked on the float), and this temperature should be reported with the specific gravity. For routine work, room temperature is satisfactory.

### **7. Microscopic Examination**

#### **a. Description**

A microscopic examination of the insoluble matter contained in urine to determine the presence of casts, blood cells, pus cells, epithelial cells, and crystals.

#### **b. Equipment and supplies**

- (1) Centrifuge, Electric.
- (2) Test Tube, 13 x 100 mm.
- (3) Slide, Microscope.
- (4) Cover Glass, Microscope Slide.
- (5) Microscope, Monocular, with Lamp.

#### **c. Procedure**

- (1) Mix the urine specimen thoroughly and transfer about 10 ml. to a test tube.

- (2) Centrifuge the tube containing the sample at moderate speed (1500 r.p.m.) for 5 minutes.
- (3) Carefully pour off most of the supernatant fluid, taking care not to disturb the sediment in the tube.
- (4) Shake the tube thoroughly to resuspend the sediment in the few drops of liquid remaining.
- (5) Place one drop of the sediment suspension on a microscope slide and cover with a cover glass.
- (6) Examine the slide with the microscope, using first the low-power objective to determine the presence of casts, epithelial cells and crystals, and then the high-dry objective to determine the presence of red blood cells and pus cells.

#### **d. Evaluation**

See illustrations of various urine sediment components (fig. 5).

*Casts and epithelial cells:* Report type and number found per low-power field.

*Crystals:* Report kind and frequency (occasional, few, many) found per low-power field.

*Red blood cells and pus cells:* Report number of each kind present per high-power field.

*Other organized sediments:* Report presence of other constituents in the urine sediment.

*Normal:* Amorphous sediments and occasional crystals and epithelial cells are found.

#### **e. Additional information**

- (1) Contaminants, such as pollen, fibers, diatoms, fungi, fat droplets, and starch granules, may be introduced into the urine specimen during or after collection and their presence should not be misinterpreted.
- (2) A thorough examination of the urine sediment under low power with subdued illumination should be performed to locate any unusual constituents before shifting to a higher power for their identification. The high-power field is of such small area that the rarer components may be missed unless a preliminary low-power scanning is performed.
- (3) Urine samples should ordinarily be examined within a few hours after collection because upon standing acid specimens may show a precipitate of urates and alkaline specimens a precipitate of

phosphates. If either of these precipitates has formed, remove the former by warming to 50° C., or the latter by slightly acidifying with dilute acetic acid before proceeding with the examination.

## **8. Other Physical Characteristics**

Careful observation of the physical characteristics of urine is a major diagnostic aid to the physician. In an emergency situation with limited laboratory facilities, the urine may be the most important indicator of disease and the most reliable means for its determination. It is therefore important that the technician report on the following physical characteristics and be alert to recognize any abnormalities.

### **a. Volume**

The volume of a single specimen is of little significance. If, however, facilities allow for the measurement over a 24-hour period, the rate should be reported in milliliters per hour.

### **b. Odor**

Normal urine has a characteristic odor, which becomes ammoniacal in urine that has begun to decompose. Various foods may cause a distinctive odor. Any putrid or other unusual odor should be described in the report.

### **c. Turbidity**

On standing, normal urine forms a cloud of mucus and cells (nubecula) floating in otherwise clear urine. This gradually settles to the bottom of the container unless the specific gravity of the urine is high. Turbidity due to abnormal sediment usually remains evenly dispersed and its presence should be reported. In addition, turbid samples should be examined microscopically (see section 7, page 15).

### **d. Color**

Normal urine is usually yellow in color, varying from almost colorless to deep amber depending upon the concentration. Abnormal color may be of varying shades, usually reddish or brownish from the presence of blood. Certain drugs being used by the patient may also cause an abnormal urine color.

**Explanation of Figure 5—  
Constituents of Urinary Sediments**

1. Hyaline casts.<sup>1</sup>
2. Hyaline and finely granular casts.<sup>2</sup>
3. Waxy (colloid) and granular casts.<sup>1</sup>
4. Granular and fatty casts.<sup>1</sup>
5. Epithelial casts.<sup>1</sup>
6. Blood casts.<sup>2</sup>
7. Mucous threads and cylindroids.<sup>2</sup>
8. Pseudocasts composed of swollen epithelial cells.<sup>1</sup>
9. *A.* Vaginal epithelium;  
*B.* Ureteral epithelium;  
*C.* Renal epithelium;  
*D.* Epithelium from pelvis of kidney;  
*E.* Spermatozoa.<sup>1</sup>
10. Squamous epithelium and pus cells.<sup>2</sup>
11. Epithelium from uretha (*B*) and bladder (*A*).<sup>2</sup>
12. Epithelium from pelvis of kidney.<sup>2</sup>
13. Leukocytes.<sup>2</sup>
14. Erythrocytes.<sup>2</sup>
15. Molds.<sup>1</sup>
16. Artefacts.<sup>1</sup>
17. Uric acid.<sup>1</sup>
18. Calcium urate.<sup>1</sup>
19. Acid ammonium urate.<sup>1</sup>
20. Calcium oxalate.<sup>2</sup>
21. Amorphous phosphates.<sup>1</sup>
22. Triple phosphates.<sup>2</sup>
23. Calcium sulfate.<sup>1</sup>
24. Leucine (round) and tyrosine (needles in tufts).<sup>1</sup>

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<sup>1</sup> After Rieder.

<sup>2</sup> After Todd and Sanford.





Figure 5.—Constituents of Urinary Sediments

# BLOOD ANALYSIS

## A. COLLECTION AND PREPARATION OF SPECIMEN

### 1. Capillary Puncture Technique

This method may be used to obtain small volumes of blood for grouping, cell counts, and other laboratory procedures requiring only a drop or two of sample. The site chosen may be either the palmar surface of a fingertip or the edge of an ear lobe. In infants the lower surface of the great toe or the heel is frequently used. The site, which should be clean and dry, is rubbed with alcohol and dried and a puncture is made with a sterile, disposable lancet or sterile, sharp needle. To prevent cross infection, the disposable lancet should be discarded and the needle heat-sterilized before reuse. The first drop of blood is wiped away; the second or succeeding drops are used for examination. Blood may be collected in a heparinized capillary tube for hematocrit, in a blood diluting pipette for cell count, or directly on a glass slide or cover slip for smears. Figure 6 illustrates these various techniques.

### 2. Venipuncture Technique

In this method the bend of the elbow is cleansed with alcohol and dried with sterile cotton or a gauze pad. A tourniquet is applied with moderate pressure to the upper arm, or an assistant may grasp the upper arm firmly if a tourniquet is not available. The patient extends his arm and clenches his fist to distend the veins. A sterile needle attached to a sterile syringe with depressed plunger is inserted into any prominent vein near the bend of the elbow. Generally the basilic or cephalic vein is selected. The needle should be about 20–22 gauge and should enter the skin about 3 mm. from the vein with the bevel at its tip uppermost. Two movements are required: one to puncture the skin and a second to seek out the vein. The size of syringe depends on the amount of blood to be drawn. After sufficient blood is drawn, remove the tourniquet, have the patient unclench his fist, withdraw the needle, place sterile alcohol-dampened gauze over the site, and ask the patient to flex his forearm immediately and hold it in this position for a few minutes. Figure 7 demonstrates the venipuncture technique.

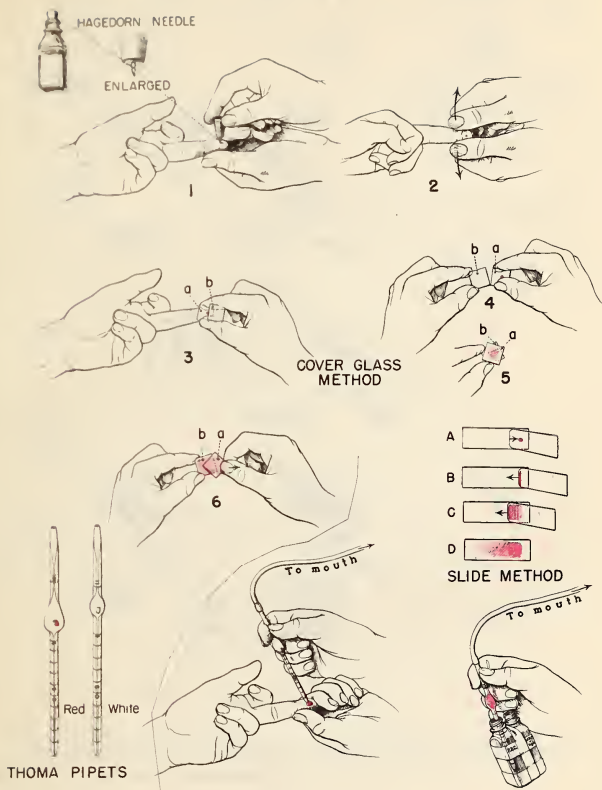


Figure 6.—Capillary Puncture Technique



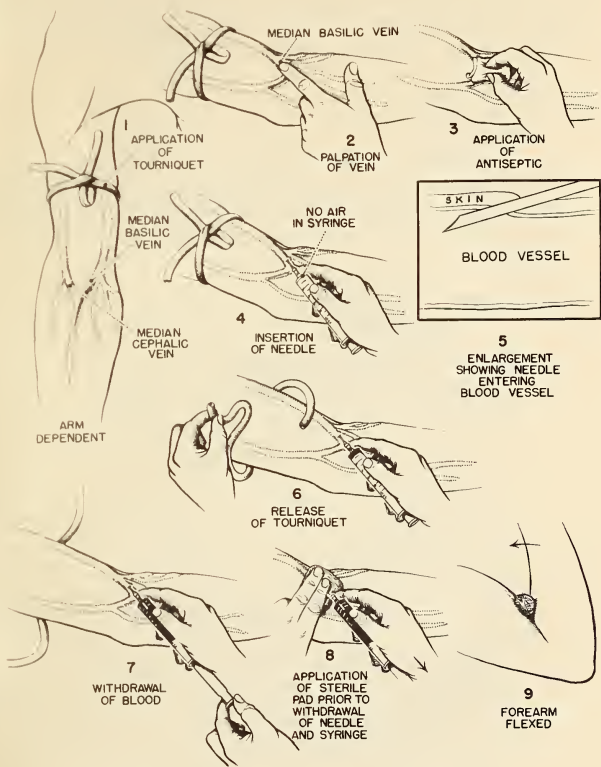


Figure 7.—Techniques of Venipuncture

The blood sample is transferred to a test tube which is then stoppered and labeled for delivery to the laboratory.

## B. TESTS

The reagents used in the tests described below should be handled carefully to avoid contamination and undue exposure to air, moisture, light, fumes, or heat. Bottles should be recapped tightly immediately after each use and

any special storage precautions stated on the labels should be strictly observed. Some of the reagents are especially subject to deterioration, and if the quality of any reagent is suspected, control tests on known samples should be performed to check its response and sensitiveness.

## **1. A-B-O Grouping**

### **a. Description**

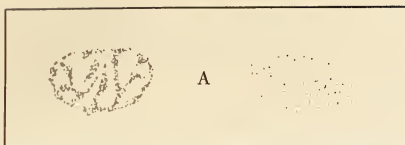
The slide method described below can be used on fresh blood, blood containing anticoagulant, saline cell suspensions or resuspended clotted blood. The use of the latter is described here. The group is determined by the reaction of the red blood cells to standard anti-A and anti-B sera.

### **b. Equipment and supplies**

- (1) Applicator, Wood.
- (2) Blood Grouping Serum, Anti-A.
- (3) Blood Grouping Serum, Anti-B.
- (4) Pencil, China-Marking, Red.
- (5) Pipette, Dropping (Medicine Dropper).
- (6) Slide, Microscope.
- (7) Microscope.
- (8) Rack, Test Tube.

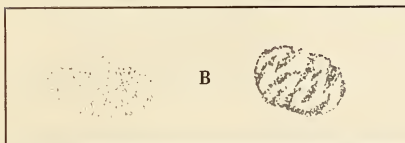
### **c. Procedure**

- (1) If the blood sample is fresh, allow to stand until clotting occurs.
- (2) Mark 1-inch squares on a microscope slide with a wax pencil and label "A" on left and "B" on right.
- (3) Thoroughly stir the clotted blood with a clean applicator stick to resuspend the cells in the blood serum.
- (4) Place a small drop of the cell suspension on each of the two squares marked on the slide.
- (5) On the left hand square place a large drop of anti-A grouping serum, and on the right hand square place a large drop of anti-B grouping serum. The drop of grouping serum should be four times the size of the drop of blood used.
- (6) Mix the cells and grouping serum with the clean end of an applicator stick for each square.
- (7) Rotate or incline the slide to keep the mixture moving for 3 minutes.



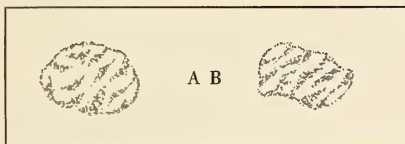
ANTI-A  
SERUM

ANTI-B  
SERUM



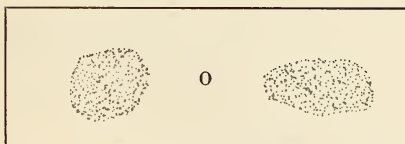
ANTI-A  
SERUM

ANTI-B  
SERUM



ANTI-A  
SERUM

ANTI-B  
SERUM



ANTI-A  
SERUM

ANTI-B  
SERUM

Figure 8.—Blood Groupings

(8) Observe for clumping of cells (agglutination) by naked eye and confirm by microscopic examination.

**Note:** *The mixture must not be warmed as this may minimize or even reverse agglutination.*

#### **d. Evaluation**

If agglutination occurs, distinct clumps will appear within a few seconds. Within 3 minutes, with continuous rotation, the clump should be 1 mm. in diameter (fig. 8). Report the blood group of the sample on the following basis:

*No agglutination on either side:* Group O.

*Agglutination with anti-A serum only:* Group A.

*Agglutination with anti-B serum only:* Group B.

*Agglutination with anti-A and anti-B sera:* Group AB.

#### **e. Additional information**

To verify group O blood for emergency use without crossmatching, a check test should be performed for each sample in one of the following ways:

(1) Use group O serum if available.

(2) Have another technician repeat test using the same anti-A and anti-B sera.

(3) Have the same technician repeat test using anti-A and anti-B sera of a different lot number.

(4) Reverse or serum typing, using known A and B cells with the donor's serum, is a valuable check if it can be performed.

**CAUTION:** *Use extreme care in handling the grouping sera to avoid any possibility of contaminating one serum with the other.*

## **2. Rh Typing**

### **a. Description**

The slide method described below can be used on resuspended clotted blood or blood containing anticoagulant. The type is determined by the reaction of the red blood cells to standard anti-Rh<sub>0</sub> typing serum.

### **b. Equipment and supplies**

(1) Applicator, Wood.

(2) Light, Extension, Trouble.

(3) Pipette, Dropping (Medicine Dropper).



(4) Slide, Microscope.

(5) Typing Serum, Anti-Rh, Anti-Rh<sub>0</sub> Blocking Type.

### **c. Procedure**

(1) If the blood sample is fresh, allow to stand until clotting occurs.

(2) Thoroughly stir the clotted blood with a clean applicator stick to resuspend the cells in the blood serum.

(3) Place a large drop of the cell suspension and a small drop of the typing serum side by side on a slide. *The drop of blood should be twice the size of the drop of typing serum used.*

(4) Mix the cell suspension and typing serum thoroughly with the clean end of an applicator stick, spreading the mixture over an area of the slide.

(5) Rotate or incline the slide to keep the mixture moving for 2 minutes. During this process keep the slide warmed to 45°–50° C. by holding it close above a lighted lamp bulb.

(6) Observe for clumping of cells (agglutination).

### **d. Evaluation**

If agglutination occurs, distinct clumps will begin to appear in about 30 seconds and the reaction will be essentially complete in 2 minutes. Report the blood type of the sample on the following basis:

*No agglutination:* Rh-negative.

*Agglutination:* Rh-positive.

### **e. Additional information**

(1) Blood specimens giving a weak or questionable reaction should be considered as Rh-positive. Blood should not be called Rh-negative unless careful inspection at the end of 2 minutes reveals no indication of agglutination. (Approximately 85 percent of the population have Rh positive blood.)

(2) A crossmatch test of Rh-negative donor blood with that of the recipient by the indirect antihuman globulin (Coombs) method is recommended if facilities for its performance are available.

## **3. Crossmatching**

### **a. Description**

A test tube method for determining the compatibility of the

recipient's serum with the donor's erythrocytes and the donor's serum with the recipient's erythrocytes, using the high-protein technique.

**b. Equipment and supplies**

- (1) Albumin, Bovine Serum, 30 percent.
- (2) Applicator, Wood.
- (3) Beaker, Laboratory, Glass, 400 ml.
- (4) Burner, Alcohol.
- (5) Centrifuge, Electric.
- (6) Pipette, Dropping (Medicine Dropper).
- (7) Sodium Chloride Tablets, USP (for normal saline), 2.25 Gm.
- (8) Test Tube, without Lip, 13 × 100 mm.
- (9) Timer, Interval.
- (10) Thermometer, Self-Indicating, minus 10° to plus 110° C.
- (11) Tripod.
- (12) Rack, Test Tube.

**c. Procedure**

- (1) Allow samples of donor's and recipient's blood to clot.
- (2) With a medicine dropper remove small specimens of supernatant serum from donor's and recipient's clotted blood samples. Place each specimen in a separate clearly marked tube.
- (3) *Major side:* Prepare a 2 percent suspension in physiological saline of cells from the donor's clotted blood sample. Into a clean test tube place one drop of 2 percent suspension of the donor's cells, two drops of the recipient's serum, and three drops of 30 percent bovine albumin.
- (4) *Minor side:* Prepare a 2 percent suspension in saline of cells from the recipient's clotted blood sample. Into a clean test tube place one drop of 2 percent suspension of the recipient's cells, two drops of the donor's serum, and three drops of 30 percent bovine albumin.
- (5) Make a preliminary check of each tube for agglutination. If clumping of the cells has already occurred in either of the tubes, incompatibility on that side is indicated and further testing of that tube is not needed.
- (6) Shake the tubes well and incubate at 37° C. for 10 minutes.

(7) Centrifuge to pack the cells firmly (2,000 r.p.m. for 2 minutes in a small radius centrifuge head; lower speeds may be optimal in larger radius heads).

(8) Gently agitate the tubes to dislodge cell packs. Observe to determine if agglutination has occurred. Apparent absence of agglutination should be verified by a microscopic examination of the cells.

#### **d. Evaluation**

Incompatibility as indicated by agglutination in this type of crossmatch may show either blood-group reaction or other antigen-antibody reaction. This test detects most incompatibilities, but certain possible reactions due to incomplete antibodies will be missed.

*Incompatibility on the major side* indicates the blood must not be given, because a serious reaction would result.

*Incompatibility on the minor side* is present when group O blood is matched with recipient blood of other groups, and when blood of group A or B is matched with AB recipients. *The major side should be compatible in these cases.*

#### **e. Additional information**

(1) The label on the donor blood must show an identifying name or number and expiration date. The label should be clearly marked to show the blood group and Rh type of the blood contained in the bottle. If the manufacturer's label is the only one available, the above information must be clearly written on it in pencil or waterproof ink.

(2) Pseudoagglutination (rouleaux formation) may be mistaken, particularly by an inexperienced worker, for true agglutination. Under the microscope the cells look like rows of stacked coins. Rouleaux formation occurs most readily when there is too great a concentration of serum or when the reading is too long delayed. The addition of a drop of saline usually disperses rouleaux formation but not true agglutination.

(3) Crossmatching is a complex procedure and should be performed only by a technician who has had training and previous experience in the performance of the test.

### **4. Hematocrit**

#### **a. Description**

A determination of the percent of the total volume of a blood

sample occupied by the red blood cells when packed by centrifugation.

#### **b. Equipment and supplies**

- (1) Centrifuge, Electric (with Hematocrit Head).
- (2) Isopropyl Alcohol.
- (3) Lancet, Finger Bleeding.
- (4) Pad, Gauze 2 x 2".
- (5) Reader, Microhematocrit Tube.
- (6) Sealing Compound.
- (7) Timer, Interval.
- (8) Tube, Capillary (heparinized).
- (9) Tube, Capillary (nonheparinized).

#### **c. Procedure**

- (1) Fill two heparinized capillary tubes about  $\frac{2}{3}$  full with fresh blood obtained from the patient's finger. Venous blood with an added anticoagulant may be used for this test if desired, in which case plain nonheparinized capillary tubes are used.
- (2) Seal one end of each tube (the end opposite from the blood) by plugging it with sealing compound or by passing it through a small flame to fuse the glass.
- (3) Place the tubes in the centrifuge head with the open ends toward the axle.
- (4) Centrifuge for 3 minutes at top speed.
- (5) A reading of the hematocrit value is obtained by measuring the height of the packed red-cell volume compared to the total volume by the use of the hematocrit reader. This gives the result directly in percent by volume.

#### **d. Evaluation**

Report the volume percent of the packed red blood cells to the nearest unit.

##### *Normal:*

Men—40% to 54%.

Women—37% to 47%.

#### **e. Additional information**

Soft rubber gaskets should be used to support the bottom of the sealed tubes in the hematocrit head of the centrifuge to prevent breakage or leakage of the tubes.

## 5. Leukocyte (White Cell) Count

### a. Description

A method for determining the number of white cells (leukocytes) per unit volume of blood by using a microscopic counting technique after diluting the blood with a fluid which hemolyzes the red cells (erythrocytes).

### b. Equipment and Supplies

- (1) Chamber, Counting, Hemacytometer.
- (2) Cover Glass, Microscope Slide, Hemacytometer.
- (3) Hydrochloric Acid, N/10.
- (4) Isopropyl Alcohol.
- (5) Microscope.
- (6) Needle, Hypodermic, 20 gauge, 1½".
- (7) Pad, Gauze, 2" x 2".
- (8) Pipette, Blood Diluting, White Corpuscle.
- (9) Pipette, Blood Diluting, Red Corpuscle.
- (10) Syringe, Luer, 10 ml., or Lancet, Finger Bleeding.

### c. Procedure

- (1) Draw fresh blood directly from a finger puncture of the patient exactly to the 0.5 mark in a white cell pipette. If desired, venous blood with an added anticoagulant may be used in this test as a substitute for fresh blood.
- (2) Wipe off any blood adhering to the outside of the pipette tip with gauze.
- (3) Draw N/10 hydrochloric acid diluting fluid into the pipette to bring the total volume to the 11.0 mark. This will provide a 1:20 dilution of the blood in the bulb of the pipette (a dilution factor of 20).
- (4) Seal off the ends of the pipette with the fingers and shake for 3 minutes to mix the contents.
- (5) Expel and discard the first four drops from the pipette.
- (6) Place the hemacytometer cover glass over the ruled platform of the counting chamber. Fill both chambers of the hemacytometer by holding the pipette with the tip at the edge of cover glass and allow a drop of diluted blood from the pipette to run by capillary attraction under the cover slip without any forcing. The drop must be large enough to cover the platform and yet not so large that it runs into the moat.

- (7) Wait for three minutes to allow the cells to settle.
- (8) Using the microscope under low power, count the number of leukocytes in the four large corner squares (each of which consists of 16 smaller squares). Begin counting at the extreme upper left of each large square and work to the right, then down to the next line and work to the left. Continue back and forth until the leukocytes in all 16 small squares within each of the large corner squares are counted. Count those cells touching dividing lines to left and above and omit those touching dividing lines to the right and below. The greatest variation between the count in each of the four large squares should not exceed 12.
- (9) Add together the number of white cells counted in all four large squares and multiply by 50.
- (10) Repeat the counting procedure, steps (8) and (9), for the second chamber and average the two results. This value is the white cell count per cubic millimeter.
- (11) If the white cell (leukocyte) count as determined above is less than 2500, repeat the entire procedure, steps (1) through (10), except in step (1), draw blood to the 1.0 mark of the white cell pipette instead of the 0.5 mark, and in step (9), use a multiplying factor of 25 instead of 50.
- (12) If the white cell (leukocyte) count is markedly elevated (as in many patients with leukemia), use the red cell pipette in place of the white cell pipette in making the dilution in step (1). If blood is drawn to the 0.5 mark with this pipette (and the diluting fluid is added to the 101 mark), the multiplying factor for use in step (9) is 500. If, in using the red cell pipette, blood is drawn to the 1.0 mark and the diluting fluid is added to the 101 mark, the multiplying factor for use in step (9) is 250.

#### **d. Evaluation**

*White blood cell (leukocyte) count:* Report the number of cells per cubic millimeter (cu. mm.) as determined above.

*Normal:* 5,000 to 10,000/cu. mm.

#### **e. Additional information**

(1) The large corner squares of the ruling on the hemacytometer are exactly 1 mm. square. The depth of the chamber is 0.1 mm. Accordingly, the general formula for determining the multiplying factor for use in step (9) of the procedure above is:

$$\frac{\text{Dilution factor}}{0.1 \times 1 \times \text{number of squares counted}}$$

(2) The pipette, hemacytometer, and cover glass must be scrupulously clean and dry before being used. In cleaning the pipette it is advisable to partially fill it with detergent solution and agitate thoroughly. Then rinse with tap water, distilled water, and acetone in order, followed by drying with a stream of air. As a substitute for acetone, alcohol followed by ether may be used. All equipment should be cleaned as soon as possible after each use.

## **6. Differential Leukocyte (White Blood Cell) Count**

### **a. Description**

A method for determining the percentages of the various types of white cells present in blood by a microscopic count of these cells in a stained blood smear.

### **b. Equipment and supplies**

- (1) Dish, Biological Staining, Glass, with Cover and Insert.
- (2) Immersion Oil.
- (3) Isopropyl Alcohol.
- (4) Lancet, Finger Bleeding.
- (5) Microscope, Monocular.
- (6) Pad, Gauze, 2" x 2".
- (7) Pipette, Dropping (Medicine Dropper).
- (8) Slide, Microscope.
- (9) Timer, Interval.
- (10) Water, Demineralized (or Distilled).
- (11) Wright's Blood Stain Kit (Consists of Tablets and Methyl Alcohol).
- (12) Paper, Filter, Qualitative.

### **c. Procedure**

(1) Transfer a small drop of fresh blood from the patient, obtained by finger puncture, to the center of one half of a clean microscope slide. (A drop of venous blood with an added anticoagulant other than oxalate may be used in this determination as a substitute for fresh blood.)

(2) Hold one end of another slide above the first at an angle of 30 to 45 degrees. Touch the drop of blood with the end of this slide so that the drop is within the angle formed. Move the upper slide slightly so that capillarity will spread the blood along the end of this slide. Then move the angled slide in the opposite direction,

spreading the blood over the first slide in a thin film. Dry the film by waving the slide in the air.

(3) Cover the blood film with 8 or 10 drops of Wright's stain solution and allow to stand 1 to 2 minutes. (Staining time will vary with each batch of Wright's staining solution and should be adjusted to obtain a distinctive coloring of the cells.)

(4) At the end of the 1- to 2-minute period, add 8 or 10 drops of demineralized or distilled water to the staining solution on the slide and allow to stand for 3 to 4 minutes until a distinctive coloring of the cells is obtained.

(5) Wash the diluted stain off the slide by flooding with demineralized or distilled water.

(6) Dry the slide by carefully blotting with filter paper and waving high above a flame.

(7) Examine under the microscope using oil immersion technique and count the number of the various types of leukocytes present by following a definite path, moving the slide up and down, and from right to left in an orderly fashion. A large area of stained film should be examined to obtain a representative count.

In general, for leukocyte counts up to 10,000 per cubic millimeter classify 100 cells. For each increase of 5000 in the total count classify an additional 100 cells up to a maximum of 500. leukocytes are to be classified as neutrophils, lymphocytes, monocytes, eosinophils, and basophils. (See figure 9 for the appearance of the various types of stained leukocytes.)

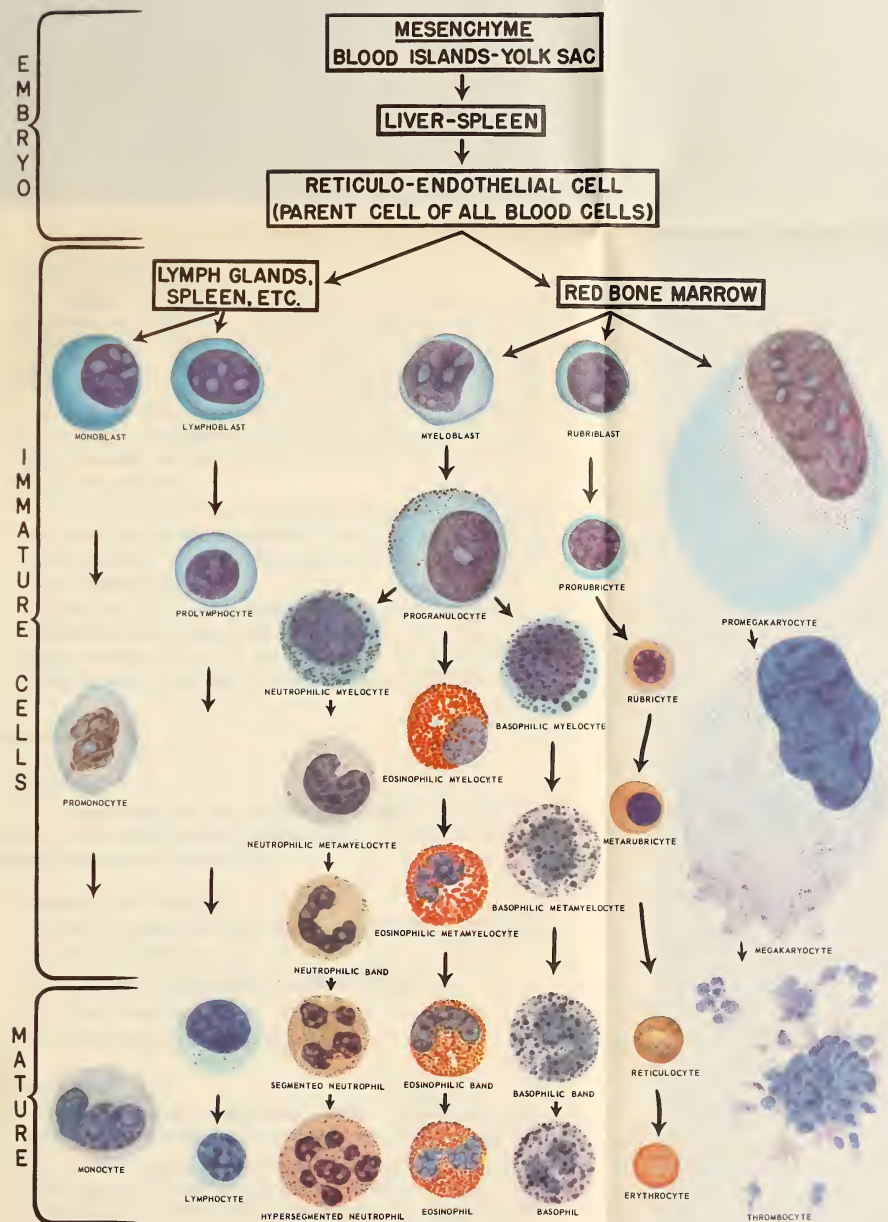
#### d. Evaluation

*Differential count:* Calculate and report the number of leukocytes of each type on both a percentage and number per cubic millimeter basis, calculating the latter from the percentage and the total leukocyte count.

*Normal (adult):*

Leukocyte	Percent	Actual number of cells per cu. mm.
Neutrophils . . . . .	50-70	3000-7000
Lymphocytes . . . . .	20-30	1000-3000
Monocytes . . . . .	2-6	100-600
Eosinophils . . . . .	1-4	50-400
Basophils . . . . .	0.25-0.5	0-50





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Figure 9.—Development of Blood Cells X 1500



#### **e. Additional information**

(1) With correct staining technique, the slide will appear copper-colored to the naked eye. Viewed microscopically, erythrocytes appear orange or buff colored; nuclei are purple; basophilic granules are dark bluish-purple; neutrophilic granules are reddish-lilac; eosinophilic granules are red; the cytoplasm of the lymphocyte is a robin egg blue and the cytoplasm of the monocyte has a faint bluish tinge. In general, a short staining time favors the eosinophilic granules, whereas a longer period favors the staining of the basophilic particles.

(2) During the microscopic counting procedure for the leukocytes, the erythrocytes should be examined for any abnormalities, including variations in size, shape, or hemoglobin content, or the presence of any abnormal inclusions.

(3) The Wright's staining solution used in this determination is prepared from the Wright's Stain Kit included in the CDEH. The tablets provided in the kit are dissolved in the methyl alcohol furnished in the proportion of 1 tablet to 10 ml. of methyl alcohol.

### **7. Total Protein**

#### **a. Description**

A color comparison test for the total protein content of blood based upon the biuret reaction in which cupric iron produces a violet color with protein in alkaline solution.

#### **b. Equipment and supplies**

- (1) Biuret Reagent Kit (Includes Reagent Powder, Test Tubes, Pipettes, and Color Chart).
- (2) Graduate, Liquid, Laboratory, Glass, 1000 ml.
- (3) Timer, Interval.
- (4) Water, Demineralized (or Distilled).

#### **c. Procedure**

- (1) Allow a sample of the patient's blood to clot and separate a portion of the cell-free serum by centrifugation or other means.
- (2) Prepare a biuret solution by dissolving the dry biuret reagent from the kit in 1000 ml. of demineralized or distilled water. (Normal saline solution may be substituted for the water if necessary.)
- (3) Fill a test tube from the kit to the calibration mark (4.0 ml.) with the biuret solution.

- (4) Fill a pipette from the kit to the calibration mark (0.1 ml.) with the patient's blood serum.
- (5) Add the contents of the pipette to the solution in the test tube by gently blowing into the pipette.
- (6) Shake the test tube gently to obtain thorough mixing and allow to stand for 15 minutes.
- (7) Using a white background about  $\frac{3}{4}$  inch behind the test tube, compare the color of the solution in the tube with the color comparison chart.

#### **d. Evaluation**

*Protein Content:* Report the protein content of the blood as *normal* or *reduced* and estimate the value to the nearest gram-percent by determining the best color match with the chart.

*Normal:* 7.5 percent.

#### **e. Additional information**

The biuret reagent solution after preparation may be stored and used as required for additional tests.

### **8. Glucose**

#### **a. Description**

A color comparison test for determining the blood sugar level based upon the reduction of an alkaline copper sulfate solution by reducing sugars.

#### **b. Equipment and supplies**

- (1) Blood Sugar Test Kit (Includes Reagent Tablets, Special Test Tubes, Filter Paper, Color Chart, and Funnel).
- (2) Pipette, Serological, 1 ml.
- (3) Rack, Test Tube.
- (4) Stopper, Bottle, Rubber.

#### **c. Procedure**

- (1) Place demineralized or distilled water in the narrow graduated test tube labeled **P** to the 1 mark (2 ml.) and add one **P** reagent tablet. While tablet **P** is dissolving, fold a sheet of the filter paper and place it dry in the funnel on the test tube labeled **S**.
- (2) After the reagent tablet **P** has dissolved, add 1 ml. of the whole blood sample to tube **P** to bring the total volume to the 2 mark. Invert and shake the contents of the tube to mix thoroughly.

(3) Immediately pour the brown mixture from the tube **P** into the filter paper over the tube **S**. Collect filtrate to the mark on the latter tube.

(4) Add one **S** reagent tablet to the filtered solution, and after the reaction ceases, shake the test tube briskly three or four times, making certain that the foam is carried down into the solution.

(5) Allow to stand for 30 seconds and then compare the color of the tube contents with the color chart.

(6) If the blood sugar level as determined above is over 200 mg. per 100 ml., repeat the entire procedure in steps (1) through (5) above, except in step (1) use 2.5 ml. of water instead of 2 ml., and in step (2) use 0.5 ml. of blood instead of 1.0 ml. In evaluating the results from this procedure, multiply the reading from the color chart by 2.

#### **d. Evaluation**

*Glucose content:* Report the findings to the nearest 50 mg. per 100 ml. by determining the best match with the color chart.

*Normal:* 70 to 120 mg. per 100 ml.

#### **e. Additional information**

(1) The **P** reagent tablets used in this test contain sulfosalicylic acid to precipitate blood proteins which would cause interference. Sodium bicarbonate is also included in their formulation to expedite dissolving.

(2) The **S** reagent tablets contain copper sulfate, citric acid, sodium carbonate and sodium hydroxide. The latter ingredient makes these tablets very hygroscopic and very irritating to the skin. They should be handled with caution.

(3) The **S** reagent tablets are especially subject to deterioration both from moisture and heat. Any tablets showing dark blue or black discoloration or poor effervescence upon dissolving may have deteriorated to such degree that incorrect results will be obtained from their use. Samples from such bottles should be checked against known glucose solutions before being used in tests.

### **9. Bilirubin**

#### **a. Description**

A color-comparison test for the presence of bilirubin in blood based upon the formation of a blue or purple colored compound with p-nitrobenzene diazonium p-toluene sulfonate.

#### **b. Equipment and supplies**

- (1) Bilirubin Test Kit (Includes Reagent Tablets and Fiber Mats).
- (2) Pipette, Dropping (Medicine Dropper).
- (3) Centrifuge, Electric.

#### **c. Procedure**

- (1) Allow a sample of the patient's blood to clot and separate a portion of the cell-free serum by centrifugation or other means.
- (2) Put two drops of the blood serum sample on a square of the asbestos-cellulose mat supplied with the kit.
- (3) Place one reagent tablet on the center of the moistened area of the mat.
- (4) Flow five drops of water over the surface of the tablet, thus causing some of the tablet to dissolve and wash onto the mat.
- (5) Observe for 30 seconds for the formation of a blue or purple color on the mat around the tablet.

#### **d. Evaluation**

*Negative:* No blue or purple color appears on the mat within 30 seconds. (A slight pink or red coloration should be interpreted as negative.)

*Positive:* A blue or purple color appears on the mat around the tablet within 30 seconds. The amount of bilirubin present in the sample is roughly proportionate to the speed and intensity of the color reaction. Report a positive result as *trace, moderate, or strongly positive*.

*Normal:* Negative.

*Sensitiveness:* A trace positive result indicates a serum bilirubin level of about 1 mg. per 100 ml.

#### **e. Additional information**

The tablets may darken from their original cream color on long storage, particularly at high temperature, but this does not necessarily mean that they are unsuitable for use.

# BACTERIOLOGY

The CDEH laboratory has a very limited capability in staining and identification techniques. It has no facilities for culturing. Staining of direct smears is limited to Loeffler's Methylene Blue and Ziehl-Neelsen Acid-Fast. If a bacteriologist is available postdisaster, the capability of the CDEH laboratory can be expanded in this area as operational conditions permit and as supplies from outside sources become available.

## A. COLLECTION AND PREPARATION OF SPECIMEN

Although a wide variety of materials can be examined microscopically by the stained smear technique, the collection of only a few kinds of samples is described here because of the limited capability of the CDEH laboratory in this field.

Pus is collected from ulcers or surface wounds by the *swab* technique, using a cotton-tipped applicator stick to pick up quantities of the exudate. The swab is then inserted in a sterile test tube for delivery to the laboratory.

Pus from subsurface infections is usually collected by aspiration with a sterile syringe and needle after first cleansing the skin around the infection with an antiseptic. The aspirated material is then expelled into a sterile test tube and the mouth of the tube closed with a loose plug of cotton.

Sputum samples are collected directly in sterile wide-mouth glass jars with suitable closures. The patient should be instructed to avoid contaminating the outside of the container when collecting the specimen. The jar is then capped for delivery to the laboratory.

To prepare a smear for staining, transfer a portion of the specimen to a microscope slide with a wire inoculating loop previously sterilized by heating in an alcohol burner flame. Spread the material over a large area in the center of the slide with the loop, adding more of the specimen as required to obtain an even film of moderate thickness. Dry the film by waving the slide, with the specimen side up, back and forth through an alcohol burner flame several times. The slide should become warm during this procedure, but not enough so to char or discolor the smear. Allow the slide to cool before staining.



## **B. STAINED SMEAR EXAMINATIONS**

### **1. Loeffler's Methylene Blue Stain**

#### **a. Description**

A general stain suitable for the morphological study of many organisms. The polychrome staining properties cause it to stain the granular portions of certain bacteria purple and the remainder of the cell body a light blue.

#### **b. Equipment and supplies**

- (1) Forceps, Cover Glass.
- (2) Microscope, Monocular, with Lamp.
- (3) Methylene Blue Staining Solution, Loeffler's.
- (4) Immersion Oil, Microscopy.
- (5) Paper, Filter, Qualitative.

#### **c. Procedure**

- (1) Cover the heat-fixed smear with the methylene blue staining solution and allow to remain for 1 or 2 minutes.
- (2) Wash the slide thoroughly by flooding with water to remove excess stain, and blot dry with filter paper.
- (3) Examine under the microscope using the oil-immersion objective.

#### **d. Evaluation**

Report the types of organisms observed, based on their morphological appearance.

### **2. Ziehl-Neelsen Acid-Fast Stain**

#### **a. Description**

A differential stain used to distinguish certain organisms possessing relatively large amounts of lipids, fatty acids, and waxes within their cells. Although resistant to staining by ordinary methods, these organisms are colored red by carbol-fuchsin in high concentration with the application of heat. Once stained, they resist decolorization by acid-alcohol, whereas other organisms are decolorized and then stained blue by a methylene blue counterstain.





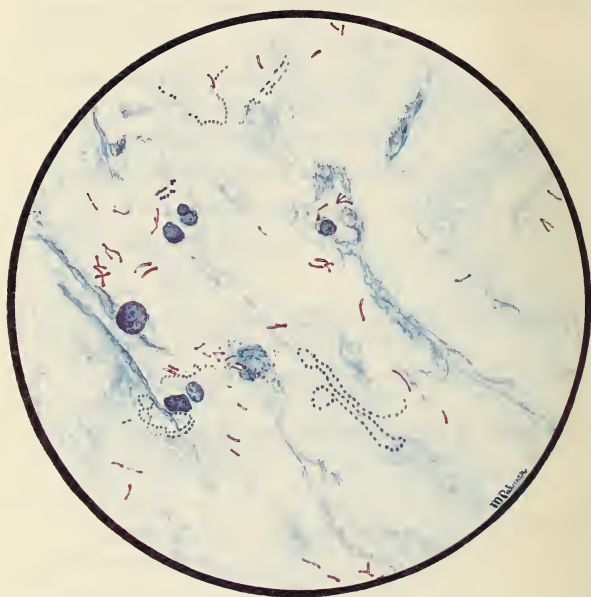


Figure 10.—Mycobacterium Tuberculosis

#### **b. Equipment and supplies**

- (1) Forceps, Cover Glass.
- (2) Microscope, Monocular, with Lamp.
- (3) Ziehl-Neelson Staining Kit.
- (4) Immersion Oil, Microscopy.
- (5) Paper, Filter, Qualitative.
- (6) Pencil, China-Marking, Red.
- (7) Burner, Alcohol.
- (8) Timer, Interval.

#### **c. Procedure**

(1) Cover the heat-fixed smear with the carbol-fuchsin solution from the staining kit, using wax pencil markings or filter paper strips at the edges of the area to be stained to keep the solution from running off the slide.

(2) Heat the slide by holding the alcohol flame (or Bunsen burner flame) for 3 to 5 minutes. The amount of heat applied should be so adjusted that the staining solution on the slide steams but does not boil.

**Note:** Do not let the slide dry; add additional staining solution as required during the heating period.

(3) Remove the excess stain with a gentle flow of water.

(4) Holding the slide at an angle, add acid-alcohol solution from the staining kit to the smear drop by drop until the washings are colorless.

(5) Remove the excess acid-alcohol decolorizer with a gentle flow of water.

(6) Cover the smear with the methylene blue counterstain from the staining kit and allow to remain for 1 minute.

(7) Remove the excess counterstain with a gentle flow of water and blot dry with filter paper.

(8) Examine under the microscope using the oil-immersion objective.

#### **d. Evaluation**

Report the types of organisms observed, based on their morphological appearance. Figure 10 shows the tubercle bacilli, *Mycobacterium tuberculosis* stained by this method.

# APPENDIX A

## LABORATORY SUPPLIES AND EQUIPMENT

### MODEL 62 CDEH

Fed. Stock No.	Item identification	Unit	Quantity
6810-753-4780	ACETONE, ACS, 1 pt (373 Gm) . . . . .	Bot.	1
6505-616-7861	ACETONE TEST TABLETS (Test for Acetone in Urine), with Color Chart, 100s.	Bot.	1
6505-000-0205	ALBUMIN, BOVINE SERUM, 30%, 5 ml. . . .	Bot.	4
6505-000-0249	ALBUMIN TEST TABLETS (Test for Albumin in Urine), 100s.	Bot.	2
.....	ALCOHOL, DENATURED—See Appendix B.		
6505-000-0250	ANTICOAGULANT SOLUTION, 1½ oz, 10% Aqueous Solution of Dipotassium Ethylenediamine Tetraacetate.	Bot.	2
6515-303-8100	APPLICATOR, WOOD, ½ x 6¼", 864s. . . . .	Pkg.	1
4320-000-0003	ASPIRATOR, PENBERTHY, with Universal Faucet Adapter.	Each	1
6640-403-0000	BASKET, TEST TUBE, Zinc-coated Steel Wire, 6 x 6 x 6".	Each	4
6640-403-3500	BEAKER, LABORATORY, Glass, 400 ml. . . .	Each	4
6505-290-6031	BILIRUBIN TEST KIT (Includes 90 Adsorption Mats and 90 Diazonium Compound Tablets).	Each	2
6810-000-0202	BIURET REAGENT POWDER to make 1,000 ml Solution (Refill for Biuret Reagent Kit 6810-000-0205).	Bot.	1
6810-000-0205	BIURET REAGENT KIT for Blood Protein (Includes Reagent Powder, 12 Test Tubes, 2 Pipettes, and Color Chart).	Each	1
6505-159-8450	BLOOD GROUPING SERUM, ANTI-A, USP, Dried, with Diluent, 75 Tests.	Pkg.	36
6505-159-8390	BLOOD GROUPING SERUM, ANTI-B, USP, Dried, with Diluent, 75 Tests.	Pkg.	36
6505-000-0253	BLOOD SUGAR TEST TABLET KIT (32 Tests): (Includes Blood Sugar Test Tablets, 6 Blood Sugar Reaction Tubes and 6 Protein Reciprocating Tubes).	Each	1
6640-408-2100	BOTTLE, URINE SPECIMEN, Glass, Graduated, without Stopper, 6-7 oz (For Stopper, see 6640-408-2105).	Each	72

Fed. Stock No.	Item identification	Unit	Quantity
7920-409-5500	BRUSH, TEST TUBE, Tufted Tip, ½" Diameter.	Each	4
6640-000-0103	CENTRIFUGE, ELECTRIC, CLINICAL MODEL, with Microhematocrit Head and Additional Head, Complete with 4 Multiple Carriers for 13 x 100 mm Test Tubes (Also see Reader, Microhematocrit, Chart Type, 6640-000-0105).	Each	1
6630-427-6850	CHAMBER, COUNTING, Blood Cells, Hemacytometer.	Each	2
6640-418-0800	CLAMP, RUBBER TUBING, Screw Regulating.	Each	2
6680-641-3205	COUNTER, RECIPROCATING, Hand-type Tally, 3 Wheel.	Each	1
6640-418-9000	COVER GLASS, MICROSCOPE SLIDE, 22 mm Square, ½ oz.	Box	12
6640-427-6875	COVER GLASS, MICROSCOPE SLIDE, Hemacytometer, Rectangular, 20 x 26 mm.	Each	12
6640-420-6000	CYLINDER, GRADUATED, LABORATORY, Mixing, with Glass Stopper, 100 ml.	Each	4
6640-299-9827	DEMINERALIZER, Water, Hand Operated . . .	Each	1
6640-422-3810	DISH, BIOLOGICAL STAINING, Glass, with Cover and insert, 20 Slide.	Each	1
5110-234-6528	FILE, TRIANGULAR, Tapered, 6" . . . . .	Each	1
6640-424-3000	FLASK, ERLLENMEYER, GLASS, 125 ml. . . .	Each	6
6640-426-0300	FORCEPS, MICROSCOPE COVER GLASS, Tweezers Type, 110 mm lg, Straight.	Each	2
6640-426-8050	FUNNEL, COMMON, Laboratory, 29° Angle, Long Stem, Glass, Smooth, 65 mm.	Each	2
6810-000-0005	HYDROCHLORIC ACID, N/10 for WBC, 1 oz.	Bot.	3
6640-299-9807	IMMERSION OIL, MICROSCOPY, 1 oz. . . .	Bot.	2
6515-000-0123	INOCULATING LOOP, Nichrome, 8" Handle, 3" Wire, 25 Gage, 3 mm Loop.	Each	1
6515-431-2890	LANCET, FINGER BLEEDING, Disposable, 100s.	Pkg.	12
6505-000-0251	LOEFFLER'S ALKALINE METHYLENE BLUE STAINING SOLUTION, 2 oz.	Bot.	1 I
6650-000-0201	MICROSCOPE, OPTICAL, MONOCULAR, 3 Objectives, Complete with Light (Light may be a separate item).	Each	1
6640-435-9000	PAPER, FILTER, QUALITATIVE, Double Acid Washed, 150 mm, 100s.	Pkg.	1
6640-559-1384	PAPER, LENS, TISSUE PAPER, Heavy-weight, 100 sheets.	Pkg.	1
6640-290-5749	PIPETTE, BLOOD DILUTING, RED CORPUSCLE, Hemacytometer, with Mouthpiece and Rubber Tubing.	Each	6
6640-427-6935	PIPETTE, BLOOD DILUTING, WHITE CORPUSCLE, Hemacytometer, with Mouth-piece and Rubber Tubing.	Each	72

Fed. Stock No.	Item identification	Unit	Quantity
6640-437-3000	PIPETTE, SEROLOGICAL, General Purpose, 0.01 ml Graduation, 1 ml.	Each	2
6640-437-4000	PIPETTE, SEROLOGICAL, Mohr Type, 0.1 ml Graduation, 5 ml.	Each	2
6640-000-0104	PROBE PIPETTE (also for Needles), 6'', 50s.	Vial	1
6640-000-0108	RACK, TEST TUBE, 72 Tubes. . . . .	Each	4
6640-000-0105	READER, MICROHEMATOCRIT TUBE, Chart Type (For use with Centrifuge 6640-000-0103).	Each	1
8030-000-0001	SEALING COMPOUND for Microhematocrit Tubes.	Pkg.	1
6640-000-0107	SLIDE, MICROSCOPE, Frosted End, 25 x 75 mm, 72s.	Box	12
8125-418-7130	STOPPER, BOTTLE, Cork, Regular Type, No. 3, 100s.	Pkg.	3
6640-441-2000	STOPPER, BOTTLE, Rubber, Solid, No. 00. . .	Each	300
6640-408-2105	STOPPER, BOTTLE, URINE SPECIMEN, Heavy Cardboard with Lift Tab, 100s.	Pkg.	12
6505-559-6859	TEST PAPER, URINE SUGAR, with Color Chart, in Dispenser, 100 Tests, 10s.	Pkg.	1
6630-000-0001	TEST PAPER, pH, with Color Chart on Dispenser, pH3-pH9 (For Refills see Test Paper, 6630-000-0002).	Each	1
6630-000-0002	TEST PAPER, pH, 15 ft. (Refill for above Dispenser, 6630-000-0001).	Roll	5
6640-443-3750	TEST TUBE, Glass, without Lip, Rated Coefficient of Expansion, $33 \times 10^{-7}$ , Wasserman, 13 x 100 mm., 12s.	Pkg.	60
6685-444-3000	THERMOMETER, SELF-INDICATING, LIQUID IN GLASS, Chemical, Minus 10° to Plus 110° C.	Each	1
6645-418-2000	TIMER, Interval, Alarm. . . . .	Each	1
6640-000-0109	TRAY, BLOOD SPECIMEN, Aluminum, 8'' x 8'' x 2''.	Each	2
6530-793-8210	TRAY, CATHETER, CRS, $8\frac{3}{4}''$ x $3\frac{5}{16}''$ x $1\frac{5}{16}''$ .	Each	2
6640-000-0110	TRIPOD, Cast Iron, I.D. $3\frac{1}{2}''$ , O.D. $4\frac{3}{4}''$ , 9'' Legs.	Each	1
6630-000-0420	TUBE, CAPILLARY, Blood Sample, Glass, Heparinized, without Rubber Bulb, 1.15 mm. I.D., 75 mm. Long, 100s.	Pkg.	2
6630-299-9837	TUBE, CAPILLARY, Blood Sample, Glass, without Rubber Bulb, 1.85 mm. I.D., 72.5 mm. Long (Nonheparinized), 100s.	Pkg.	12
4720-142-2368	TUBE, LABORATORY, Pure Gum Natural Rubber, Black, for Pressure, $\frac{3}{16}''$ I.D., $\frac{3}{16}''$ Wall, 12'.	Each	1
4720-141-9080	TUBE, LATEX, Translucent, for Tourniquets, $\frac{3}{16}''$ I.D., $\frac{3}{32}''$ Wall.	Foot	6
4720-141-9067	TUBE, RUBBER, RED, for Drainage and Laboratory Use, $\frac{1}{4}''$ I.D., $\frac{1}{16}''$ Wall.	Foot	350

Fed. Stock No.	Item identification	Unit	Quan- tity
6505-299-8624	TYPING SERUM, ANTI-Rh, USP, Anti-Rh <sub>o</sub> , Blocking Type, with Diluent, Dried (Equivalent to 3 ml.), 75 Tests.	Pkg.	36
6630-447-0000	URINOMETER, Squibb Type, with Glass Float and Cylinder.	Each	2
.....	WATER, DEMINERALIZED (Prepare by using Demineralizer, Water, Hand Operated.)		
.....	WATER, DISTILLED (Obtain locally).		
6505-149-6010	WRIGHT'S BLOOD STAIN TABLETS with Methyl Alcohol (Set consists of 12 Tablets and 4 Bottles Methanol).	Pkg.	2
6810-281-1865	XYLENE, ACS (Xylol), 1 lb. (453.6 Gm).....	Bot.	1
6505-000-0255	ZIEHL-NEELEN STAINING KIT for TBC Smears (Kit includes 2 oz. each of Methylene Blue, Carbol-Fuchsin and Acid Alcohol).	Each	1

## APPENDIX B

### OTHER CDEH SUPPLIES AND EQUIPMENT IN MODEL 62 CDEH USED IN THE LABORATORY

The following items, used basically in other departments of the hospital, are also needed in the laboratory. Laboratory personnel must obtain a suitable supply of these items from central supply section or the pharmacy when setting up the laboratory.

Federal Stock Number	Item Identification
Pharmaceuticals	
6810-000-0207	ALCOHOL, DENATURED, Special Denatured Alcohol, 23 H, 1 qt. (Primarily for use with Burner, Alcohol, 6640-410-2820, Sec. II).
6505-261-7256	ISOPROPYL ALCOHOL, NF, 1 qt.
6505-153-8708	SODIUM CHLORIDE TABLETS, USP (for Normal Saline), 2.25 Gm. (34.7 gr.), 100s.
Hospital Supplies and Equipment	
6530-406-0150	BOTTLE, SCREW CAP, Prescription, with Plastic Cap, 2 oz. 72s.
6640-410-2820	BURNER, ALCOHOL, Metal, Self-Generating, Barthel Type, 8 oz. Capacity (For Alcohol see 6810-000-0207, Sec. I).
6640-427-5250	GRADUATE, LIQUID, LABORATORY, Conical, Glass, without Handle, 1000 ml.
6530-422-8120	PIPETTE, DROPPING (Medicine Dropper), not Graduated, Glass, Rubber Bulb, 12s.
Surgical Supplies and Equipment	
6515-303-8250	APPLICATOR, WOOD, Cotton Tipped End, $\frac{1}{12}$ " x 6", 100s.
6515-349-3400	NEEDLE, HYPODERMIC, Luer Lock, Regular Bevel, 20 Gauge, $1\frac{1}{2}$ ", 12s.
6515-349-4400	NEEDLE, HYPODERMIC, Luer Lock, Regular Bevel, 22 Gauge, 1", 12s.
6515-365-1820	SCISSORS, GENERAL SURGICAL, Straight, One Point Sharp, $5\frac{1}{2}$ ".
6515-380-4100	SYRINGE, LUER, General Purpose, Small Tip, Glass, Graduated in $\frac{1}{5}$ ml. Intervals, 10 ml.



Federal Stock Number	Item Identification
<b>Surgical Dressing and Textile Products</b>	
6510-203-5500	ADHESIVE PLASTER, SURGICAL, 12'' x 10 yds., Cut in As-sorted Widths: four ½''; three 1''; two 2''; one 3''.
6510-559-3221	PAD, GAUZE, 12 Ply, Surgical, 2 x 2'', 100s.
7210-205-3087	TOWEL, HAND, Plain Bleached Huck, 17'' x 36''.
<b>Hospital Records and Office Supplies</b>	
7530-261-3807	CARD SET, GUIDE, FILE, ALPHABETICAL GUIDES A-Z, ½'' Cut, 4'' x 6''.
7520-286-6958	CASE, FILING AND TRANSFER, Binders' Board with Follower Block and Cover, 10'' Capacity, for 4'' x 6'' Forms.
7530-178-8405	LABEL, GUMMED, Blank, Oblong, with Red Border, 2½'' x 1½'', No. 201, 30s.
7530-239-8479	PAD, WRITING PAPER, White, Unruled, 5'' x 8'', 100 sheets, 12s.
7510-174-3205	PENCIL, CHINA-MARKING, Red, 12s.
7510-285-5829	PENCIL, COPYING, INDELIBLE, with Mouthpiece, Medium.
7540-634-4132	REPORT FORM, HEMATOLOGY (Standard Form 514-B), 2 Part, 500s.
7540-634-4154	REPORT FORM, MISCELLANEOUS (Standard Form 514-M), 2 Part, 500s.
7540-634-4130	REPORT FORM, URINALYSIS (Standard Form 514-A), 2 Part, 500s.
<b>Water Supplies</b>	
6640-000-0111	STILL, WATER, 2-gal. per hr. Capacity, Gasoline Burner Operated.
<b>Electrical Supplies and Equipment</b>	
6230-000-0001	LIGHT, EXTENSION, TROUBLE, with Guard, Extension Cord, Reflector, and 2 Female Receptacles, 16 Gauge, 50'.
<b>Maintenance and Housekeeping Supplies</b>	
9110-000-0001	ALCOHOL, SOLIDIFIED, 2½ oz. (For use with Stove, One-Burner, Sterno. 7310-000-0004).
9920-174-3194	MATCHES, SAFETY, 20 Matches to Book, 50 Books to Box.
7310-000-0004	STOVE, ONE-BURNER, Alcohol, Solidified (Sterno Stove) (For Alcohol see 9110-000-0001).

## APPENDIX C

### USE AND CARE OF THE MICROSCOPE

A microscope is included with the CDEH laboratory equipment for use in certain blood and urine tests and for bacteriological work. A number of different makes and models of microscopes are included in the individual CDEH. All microscopes are of the monocular type, equipped with a revolving nose piece and three objectives, one or more eyepieces, a movable stage, a substage condenser, and a microscope lamp. The lamp may be packed with the microscope or as a separate item. A detailed instruction book is packed with each microscope.

Figure 11 shows a typical microscope with the various parts labeled for identification. In addition to the parts shown, each microscope has a movable stage attachment, of either the mechanical rack and pinion type or the glide type, plus an external lamp for illumination.

The specimen to be examined is first mounted on a slide and overlaid with a cover glass as directed in the procedure for the specific test being performed. The slide is then placed on the stage of the microscope and held in place by stage clips or other device, with the specimen centered over the stage opening. The low-power objective (the shortest of the three objectives) is rotated to line up with the microscope tube and an approximate focus is obtained by rotating the coarse adjustment. Use the plane mirror to reflect light from the lamp (placed a few inches away) through the condenser and specimen while focusing. Center the area of the specimen to be examined in the field and shift to the proper higher power objective by rotating the nose piece, unless a low power examination is called for.

Adjust the illumination by focusing the condenser up or down, by opening or closing the iris diaphragm, and by moving the lamp or mirror. Obtaining proper illumination is a matter of technique which improves with practice. Best illumination is achieved when the following two conditions are met:

1. The field of view is evenly illuminated.
2. The upper lens of the objective is just filled with light when observed after removing the eyepiece from the microscope tube.

In using the low-power objective, the plane mirror and condenser can be used, or the condenser can be swung out of the optical path (or removed)

and the plane or concave mirror employed, whichever is found to be most suitable for the specimen under study. With the low-power objective, daylight from a north window can be used as a source of illumination (instead of the lamp) if desired.

For the medium-power (high-dry) objective, the condenser must be used to completely fill the field with light. Either the plane or concave mirror can be employed depending upon the type of light source and the amount of light required.

When using the high-power (oil-immersion) objective, the lamp, condenser, and plane mirror must be used. A drop of special immersion oil must be placed between the objective and cover slip. The lighting for the high-power objective can be further improved (if required) by using immersion oil between the slide and the top lens of the condenser.

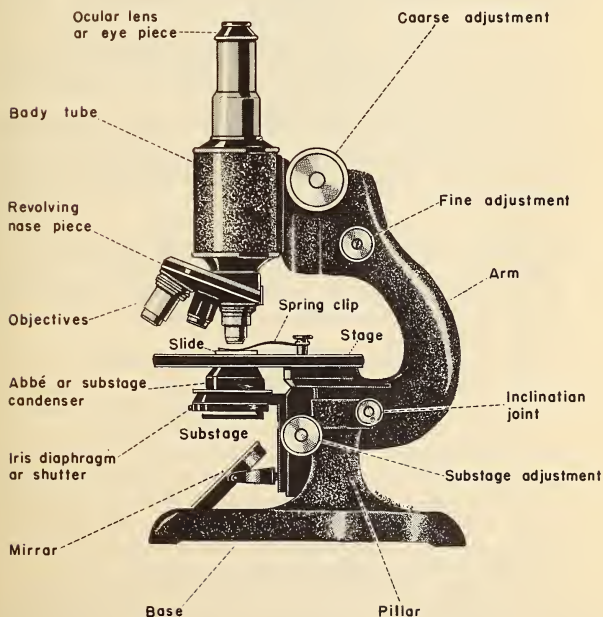


Figure 11.—The Microscope

The final adjustment of lighting can be made by using suitable filters in the lamp or condenser, after which the final adjustment of the focus is accomplished by rotating the fine adjustment knob on the microscope.

In the handling and use of the microscope, certain special techniques should be employed and certain precautions must be observed in order to obtain maximum usefulness from the instrument and to insure its continued functioning. The following suggestions are offered for those who have had limited previous experience in working with this instrument:

1. The microscope is a delicate optical instrument. It should be handled carefully. Avoid jarring and sudden shocks.
2. When moving the microscope, pick it up by the arm, never by any other part. Use the other hand to support the base while carrying the instrument.
3. Keep the microscope covered when not in use. Keep the eyepiece and objectives in the microscope to prevent dust from entering the tube. Store the microscope in a cool, dry place.
4. Clean lenses only with lens tissue or a soft lint-free cloth, after a preliminary dusting to remove coarse particles. Lens tissue may be moistened with distilled water or xylene, if necessary. Xylene is particularly useful for removing grease or dried immersion oil, but it should be used sparingly and as quickly as possible because it will dissolve the lens mounting cement in some microscopes.
5. Never disassemble any of the lens systems. Clean only the exposed surfaces of the ocular, objectives, condenser, and mirror systems, and these only when necessary. Do not touch the lens surfaces with the fingers.
6. Always use a cover glass over a specimen on a slide.
7. Do not hold the coarse adjustment knob while rotating the fine adjustment knob.
8. Always start the examination of a specimen with the low-power objective to obtain centering and approximate focus before shifting to a higher power objective.
9. When focusing with either of the two high-power objectives, be careful not to lower the objective so that it strikes the slide.
10. In using the oil immersion objective, the usual procedure is to apply a small amount of special immersion oil or cedarwood oil to the bottom lens of the objective. Lower the objective carefully with the coarse adjustment until contact is made between the oil and the slide, as indicated by a flash of light illuminating the oil. (The oil may be placed on the cover slip instead of the lens if preferred.) Then obtain the final focus with the fine adjustment only. Clean all traces of oil from the lens with lens tissue immediately after the completion of work.
11. The substage diaphragm is not intended for the control of the intensity of illumination. If the field is too bright, use one or more neutral (uncolored) filters in the light path or move the lamp away from the microscope.

## APPENDIX D

### OTHER PUBLICATIONS ON THE CIVIL DEFENSE EMERGENCY HOSPITAL

*Establishing the Civil Defense Emergency Hospital (F-1).*

*X-ray Section of the Civil Defense Emergency Hospital (F-2).*

*Central Supply Section of the Civil Defense Emergency Hospital (F-3).*

*Operation of Generators in the Civil Defense Emergency Hospital (F-5).*

*Checklist for Developing A Civil Defense Emergency Hospital Utilization Plan.*

The above publications are available, upon request, from your State Health Department, civil defense office, or Division of Health Mobilization, Public Health Service, Washington, D.C., 20201.











Publications in the Health Mobilization Series are keyed by the following subject categories:

A—Emergency Health Service Planning

B—Environmental Health

C—Medical Care and Treatment

D—Training

E—Health Resources Evaluation

F—Civil Defense Emergency Hospitals

G—Health Facilities

H—Supplies and Equipment

I—Health Manpower

J—Public Water Supply

Public Health Service  
Division of Chronic Diseases  
100, Cst. Reg. III Bldg.  
Washington, D.C.

Public Health Service  
Publication No. 1071 F-4

